The effect of soaking in water and of aëration on the growth of Zea Mays

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(WITH FIVE TEXT FIGURES)

The first of these experiments was to ascertain the best length of time required to soak the grains of *Zea Mays* for growth to begin. The grains we used were one year old. Some experiments of this kind have been made,* but since the time varies to some extent in the grains of *Zea Mays* of different kinds and ages, it was desired to determine it for the material at our disposal. The second object was to determine the effects of aëration on the seedlings of *Zea Mays* under different conditions.

1. Experiments in soaking grains of Zea Mays

a. *Time required for maximum growth*

In order to ascertain the proper length of time to soak *Zea Mays* for maximum growth the grains were kept in tap water for varying lengths of time, and then in wet sawdust long enough to total forty-eight hours. The experiments were kept under normal growing conditions.

Tables I–IV give the average length of growth of five grains. The unsoaked grain used as a control made no external growth in any of the experiments.

TABLE I

Hours soaked	Growth in mm.
16	8.6
17	10.8
19	12.6
21	10.2
23	4.6
25	3.0

TABLE II

Hours soaked	Growth in mm.
6	5.2
8	11.2
10	11.5
12	13.4
14	11.6
24	4.8

* See Burt, G. J., Biggar, H. H., & Trout, C. E. The rag-doll seed tester. U. S. Dept. Agr. Farm. Bull. 948: 1–7. *f. 1–5.* 1918. On page 4 the writers state, "it is best not to soak for more than 10 hours." We have found, however, that 12 hours is the optimum length of time with our material.

TABLE III

Hours soaked	Growth in mm.
12	26.6
15	20.4
17.5	15.4
20.5	7.2
22.5	10.2

TABLE IV

Hours soaked	Growth in mm.
1.25	0.6
3.25	2.4
5.5	4.6

The results of the above series show that the amount of time for soaking the grains of *Zea Mays* for good growth ranges from about 8–14 hours with the optimum length of time near 12 hours. The series shown in TABLE I, however, indicates 17–21 hours as the best, but this discrepancy was due to a variation in temperature.

b. *Growth in sawdust and soil compared*

It is interesting to note the effect of the use of damp sawdust to germinate *Zea Mays*, as compared with damp soil. TABLE V gives the results with using grains soaked 11 hours in tap water and remaining in the soil (or sawdust) for 37 hours longer.

TABLE V

	Unsoaked Seed	Soaked Seed
Soil	8 mm. (av.* of 24 grains)	9.4 mm. (av. of 22 grains)
Sawdust	3.1 mm. (av. of 26 grains)	9.3 mm. (av. of 25 grains)

The soaked grains showed a difference of 0.1 mm. in favor of the soil; while the unsoaked grains in the soil showed a gain of 258 per cent in favor of the soil. That was due probably to the sawdust having a greater tendency to dry out than the soil. The grains probably absorbed the moisture more readily from the soil as its particles were smaller, thus tending to establish a better condition for absorption.

c. *Effect of puncturing the coats of the grains*

The large end of the grain was punctured, after which it was soaked for 10.25 hours and left in sawdust long enough to make 48 hours. The average growth of 25 punctured grains was only 11.7 mm., due to air which was engulfed or held for a long time in the small opening made by the puncture; 11 had a growth of 10 mm. or more; 7 with a growth of less than 10 mm., and 7 without

* "Av.," wherever used in this paper, stands for "average."

visible growth. The control of 25 grains gave an average growth of 19 mm., ranging in length from 3 mm. to 44 mm. Of these 18 made a growth of 10 mm. or more, and the remaining 7 made a growth of less than 10 mm. This shows that when *Zea Mays* is soaked, puncturing the coats does not increase the growth if the opening is blocked by air.* But when unsoaked grains are used and they are in wet sawdust different results are obtained, as is shown by the following experiment.

The average growth of the 24 punctured unsoaked grains was 1.3 mm.; the growth measurements of 7 were as follows, in millimeters: 10, 6, 6, 7, 2, 1, 1, with 17 grains showing no growth. The 24 unpunctured grains gave an average of only 0.3 mm., with 21 having no growth and others ranging in length as follows: 4 mm., 2 mm., and 1 mm. This indicates that when there is no interference from air, moisture is absorbed more regularly and that puncturing of the coat materially increases the growth.

The study of the effect of punctures in different parts of the *Zea Mays* grain was next investigated. In the first experiment the coats were punctured on the opposite side of the grain from the embryo, with the following results. The unsoaked grains were planted in damp sawdust and left for 24 hours. The average growth of 25 grains was 7.04 mm. 2 seedlings were over 40 mm. long, 3 were 15–18 mm. long, 11 were less than 7 mm. long, and 9 showed no growth. The average of 24 unpunctured grains was 3.6 mm., 3 were over 10 mm. long, 7 were less than 10 mm. in length, and 14 showing no growth.

A small puncture was then made in unsoaked grains over the embryo with the fol owing results: the average growth of 25 grains was 3.24 mm. The length of the growing seedlings was as follows, in millimeters: 25, 19, 14, 10, 7, 3, 2 and 1; while 17 showed no growth. The average growth of 25 unpunctured grains was 1.88 mm. The length of the growing seedlings was as follows, in millimeters: 17, 14, 10, 3, 2, 1; while 19 showed no growth.

The removal of part of the coats from different parts of the grain was made with the following results:

* See de Vries, H. Über künstliche Beschleunigung der Wasseraufnahme in Samen durch Druck. Biol. Centralbl. 35: 161–176. 1915.

1. The seed coat was removed from the large end of the unsoaked *Zea Mays* grain which resulted as follows after 60 hours growth. The average growth of 25 grains was 12 mm. The lengths of the seedlings in millimeters was as follows, 63, 57, 27, 27, 19, 18, 15, 13, 11, 9, 7, 4, 3, 3, 2, 2, 1; while 8 showed no growth. The average growth of 25 seedlings with coats intact was 4.6 mm. The length of these seedlings was as follows, also in millimeters: 36, 26, 20, 10, 9, 8, 2, 3, 1, 1, 1; while 14 showed no growth.

2. The coats were removed from the embryo side of the unsoaked grain and the grain left in the sawdust for 43 hours. The average length of growth of 25 grains was 1.4 mm. The lengths of the seedlings was as follows in millimeters: 17, 12, 4, 3, 2, 1, 1, 1; while 17 showed no growth. The average of 25 grains with intact coats was 0.04 mm., since the only grain germinating made a growth of 1 mm.

3. The coats were removed from one side of *Zea Mays* grains and after 48 hours they showed the following growth. The average growth of 22 grains was 1.9 mm.; the lengths attained by the seedlings were as follows in millimeters: 12, 8, 7, 4, 2, 2, 2, 2, 1, 1, 1, 1; while 10 had no growth. The grains from which the coats had not been partially removed did not make growth enough to break through the coats. These experiments show the decided advantage a grain of *Zea Mays* has when parts of the coats are removed and show that the absorption of water by the endosperm takes place under such circumstances much more rapidly. In this connection it is interesting to consult the paper of A. J. Ewart and Jean White on "The Longevity of Seeds."*

II. EFFECT OF AËRATION ON THE GROWTH OF ZEA MAYS

This experiment was conducted for the purpose of ascertaining the effect of aëration on the growth of *Zea Mays*. In water cultures, as commonly conducted, the factor of aëration is one that is generally neglected. The plants were grown in most cases as water cultures according to the formula as given by J. Sachs. The cylinders used had a capacity of 1.5 liters and the solution was changed at frequent intervals. The aëration apparatus used in one set of experiments was that of Kekulé, as described by Ostwald. The other piece of apparatus used for aëration was the Bunsen filter pump.

Sachs was the first to experiment with an aëration apparatus on water cultures. This apparatus he illustrates on page 268 and describes on page 269 of his Vorlesungen über Pflanzenphysiologie. The apparatus of Sachs is simple but requires more or less attention, while either of the pieces of the apparatus used for aëration of the cultures mentioned in this paper can be regulated and left for long periods of time to run safely and regularly

* Proc. Roy. Soc. Victoria **21**: 1–203. 1908.

† See Beals, C. C. The effect of aeration on the roots of *Zea Mays*—I. Proc. Indiana Acad. Sci. **1917**: 177–180. *f. 1–3.* 1918.

‡ Manual of physico-chemical measurements 189. 1894.

without further attention. The Kekulé apparatus as described by Ostwald is very convenient for aëration. The glass tube extends to near the bottom of the cylinder to aërate the solution as perfectly as possible. The resistance can be varied by raising and lowering the tube in the cylinder, and the opening of the tube is slightly contracted to form smaller air bubbles. Care, however, must be used or the resistance will be so great that the water pressure will not force the air bubbles through with sufficient rapidity.

The Bunsen filter pump affords a greater quantity of air than the Kekulé apparatus. From the Bunsen filter pump apparatus numerous lateral tubes can be attached to a main air pipe leading from the pump so that fifteen to twenty cultures can be aërated at once. The supply of air to the individual water cultures is regulated by pinch cocks on the connecting rubber tubes and by raising or lowering these in the solutions.

A short paper appeared by Free* on the aëration of buckwheat, in 1917, in which it was found "that the degree of aeration of the culture solution is without important influence." Stiles and Jörgensen obtained the same result with buckwheat as Free.

As the columns of air carried down the tube are much larger and of much greater volume than the drops of water, it was found upon measurement that 3.5 times as much air was passed through the solution by the Kekulé apparatus as the volume of water required to convey the air. About .100 c.c. of air were passed through the solution per minute.

TABLE VI

Number of days	Aërated	Non-aërated
2	2.8 c.c.	1.9 c.c.
3	5.9 "	4.7 "
6	14.5 "	12.0 "
8	25.0 "	23.0 "
11	28.0 "	24.0 "
15	37.0 "	33.0 "
20	47.0 "	37.0 "
26	65.0 "	46.0 "

* The effect of aeration on the growth of buckwheat in water-cultures. Johns Hopkins Univ. Circ. 293: 198, 199. 1917.

† Observations on the influence of aeration of the nutrient solution in water culture experiments, with some remarks on the water culture method. New Phytol. **16**: 181–197. 1917.

TABLE VI gives the height of the plants at different stages of growth where 100 c.c. of air per minute is used.

After three months' growth under as nearly normal growing conditions as possible, the plants were removed and the amount of ash ascertained. The ash of the aërated plant including the roots were 2.181 grams while the ash of the non-aërated plant weighed 1.303 grams. In another experiment after two months' growth

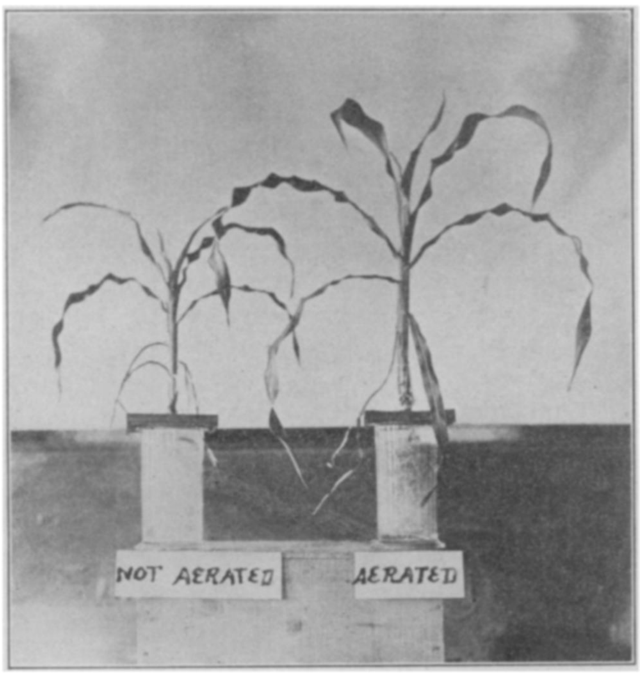

FIG. 1. Non-aërated and aërated specimens of *Zea Mays*.

the ash of the aërated plant (including the roots) weighed 1.855 grams, while the non-aërated plant weighed 0.65 grams or almost three to one in favor of the aëration. The difference is well shown in FIG. 1. The same experiment was performed a number of times, the aërated plant always showing the same marked improvement over the non-aërated one.

Another experiment, shown by FIG. 2, was performed using five cylinders. The air was driven through these cultures by means of a Bunsen Pump apparatus. The culture *A* received

1 liter of air; *B*, 3 liters; *C*, 92 liters; *D*, 120 liters; and *E* 144 liters in 24 hours. The figure shows that the plants made better growth when an increased amount of air was used.

The temperature of the culture solution was often too high for good growth. FIG. 3, *A*, is a culture grown without aërating or having the temperature lowered to a favorable point. *B* is a culture which was aërated with about 48 liters of air in 24 hours, but whose culture solution was above a favorable temperature. *C* is a

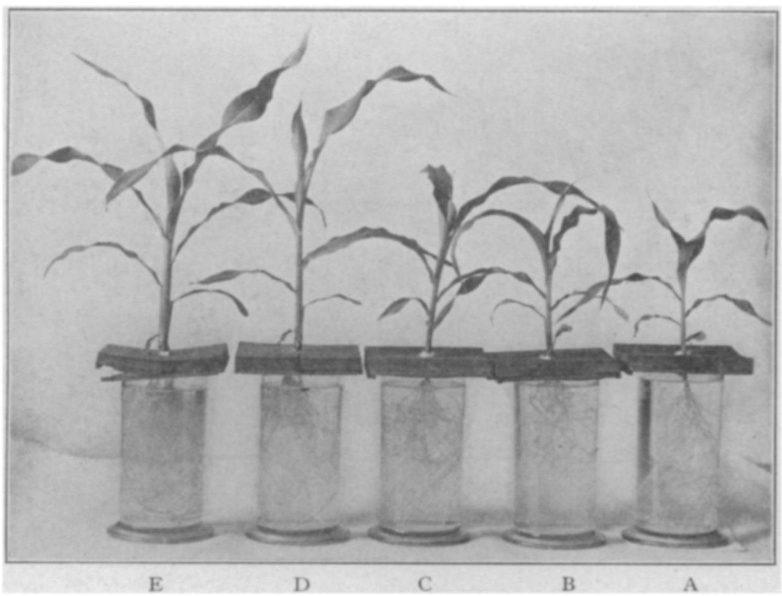

FIG. 2. Effects produced by different quantities of air passed through the culture solution. Reading from right to left: *A* received 1 liter in 24 hours; *B*, 3 liters; *C*, 92 liters; *D*, 120 liters; and *E*, 144 liters.

culture which was aërated with about 48 liters of air in 24 hours and which in addition had the temperature of the culture solution lowered to a favorable point. The temperature in the culture solution of *C* was controlled by placing the glass cylinder containing the roots in a large vessel and allowing a sufficient quantity of water of the desired temperature to flow around it. It is at once seen from the photograph, FIG. 3, that *B* has grown much better than *A*, and that *C* is much larger and has grown faster than *B*. All the cultures were started at the same time and all

the other conditions, except those of arëation and temperature, were always the same.

FIGS. 4 and 5 show parts of cross-sections of the roots of *Zea Mays* grown in water cultures. FIG. 4 is part of a cross-section of a root which had been aërated, and FIG. 5 is the same of a non-aërated root. It will be noticed that the intercellular spaces in the non-aërated root are appreciably larger.

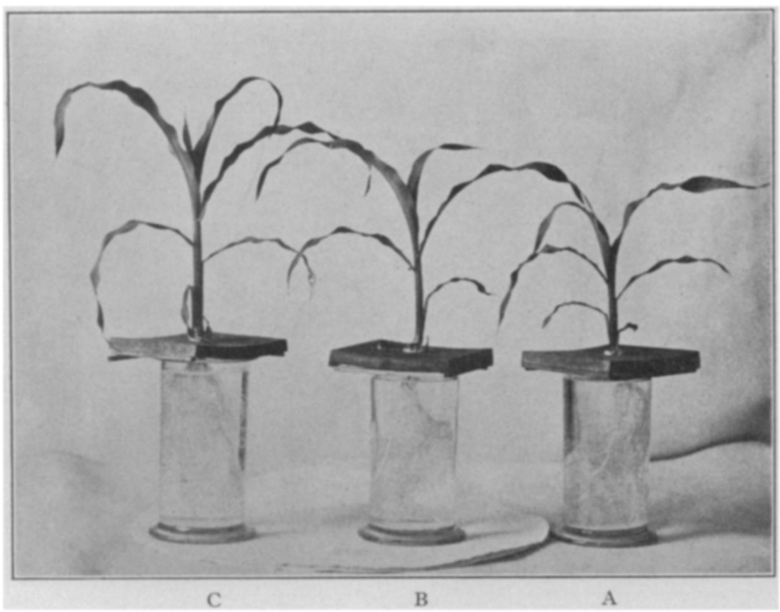

FIG. 3. Effects of temperature and aëration. Reading from right to left: *A* was not aërated and was exposed to too high a temperature; *B* was aërated with about 48 liters of air in 24 hours, but was exposed to too high a temperature; *C* was aërated with about 48 liters of air in 24 hours and was exposed to a lower and more favorable temperature.

Specimens of *Zea Mays* sometimes suffer for lack of aëration under supposedly normal conditions in the soil. To show this point the following experiment was performed. A large bottle was cut off near the bottom, a perforated rubber stopper was inserted in the neck and a glass tube put through this for the admission of air. The bottle was inverted, about 1 cm. in depth of melted paraffin poured over the cork to exclude the air and the lower part filled with wet sphagnum and this covered over with

moist earth to a depth of about two decimeters. A young *Zea Mays* plant was placed in the soil and melted paraffine was poured over the surface after placing a small short paper cylinder around the plant. The paraffine caused all the air from the aërating apparatus to come out around the plant and not escape around the sides of the bottle or elsewhere before coming in contact with the roots. The bottle was supported on a ring-stand and the small glass tube attached to the aërating apparatus. The apparatus offered practically no resistance to the air supply as was shown by a

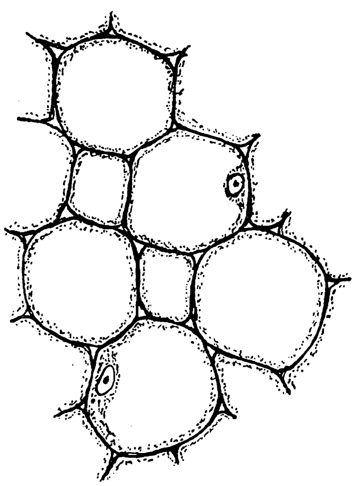

FIG. 4.

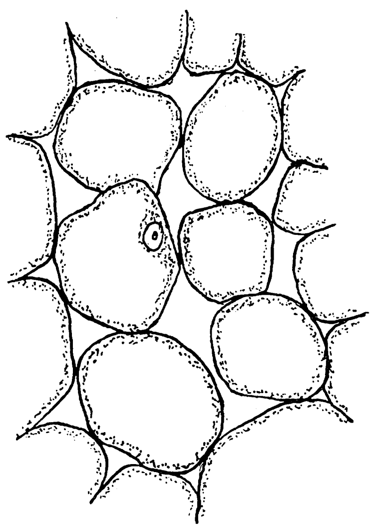

FIG. 5.

FIG. 4. Part of a cross section of a root of *Zea Mays* grown in an aërated water culture. FIG. 5. Part of a similar section of a root from a non-aërated water culture.

manometer. A control plant was planted in a pot and set on the bench near by. The aërated plant in this case made almost as great a gain over the non-aërated as the ones so treated in the water cultures, showing that the increase was due to the presence of a greater amount of air. All the other conditions except that of aëration were the same for both specimens in this experiment.

SUMMARY

1. The best length of time for soaking the grains of *Zea Mays* used in these experiments was 12 hours.

2. Puncturing the coats of the grains or the removal of a portion of the coats accelerates, as would be expected, the germination under proper conditions.

3. Too long soaking materially retards the growth for a time.

4. Aërating the culture solution accelerates the growth.

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